

Figure 1

## Sequence alignment of mouse Serca 1, 2 and 3 protein.

5	Serca1a	1	MEAAHSKSTEECLSYFGVSETTGLTPDQVKRHLEKYGNELPAEEGKSLWELVVEQFEDL
	Serca2a	1	..N..T.TV..V.GH...N.S...SLE...KLK.RW.S.....T.L...I.....
	Serca2b	1	..N..T.TV..V.GH...N.S...SLE...KLK.RW.S.....T.L...I.....
	Serca3a	1	..E..LL.AADV.RR.S.TAEG..SLE..TDAR.R.....T.....
	Serca3b	1	..E..LL.AADV.RR.S.TAEG..SLE..TDAR.R.....T.....
10	Serca3c	1	..E..LL.AADV.RR.S.TAEG..SLE..TDAR.R.....T.....
	Serca1a	61	LVRILLLAACISFVLAWFEEGEETVTAFVEPFVILLILIANAIVGVWQERNAENAIEALK
	Serca2a	61	.....I.....V.....
	Serca2b	61	.....I.....V.....
15	Serca3a	61	.....LV.....T.....L.M...V.....S.....
	Serca3b	61	.....LV.....T.....L.M...V.....S.....
	Serca3c	61	.....LV.....T.....L.M...V.....S.....
	Serca1a	121	EYEPENGKVKYRADRKSVQRIKARDIVPGDIVEVAVGDKVPADIRILSIKSTTLRVDQSIL
20	Serca2a	121	.....Q.....K.....I.....LT.....
	Serca2b	121	.....Q.....K.....I.....LT.....
	Serca3a	121	.....I.S...G...R.....L.LIE.....
	Serca3b	121	.....I.S...G...R.....L.LIE.....
	Serca3c	121	.....I.S...G...R.....L.LIE.....
25	Serca1a	181	TGESVSVIKHTDPVPDPRAVNQDKKNMLFSGTNIAAGKAVGIVATTGVSTEIGKIRDQMA
	Serca2a	181	.....M.V.VA...N.....E.V
	Serca2b	181	.....M.V.VA...N.....E.V
	Serca3a	181	.....T...AI.....S...L.VAVA..LQ..L...S...
30	Serca3b	181	.....T...AI.....S...L.VAVA..LQ..L...S...
	Serca3c	181	.....T...AI.....S...L.VAVA..LQ..L...S...
	Serca1a	241	ATEQDKTPLOQKLDEFGEQLSKVISLICVAVWLINIGHFNDPVHGGSWFRGAIYYFKIAV
35	Serca2a	241	...ER.....I...I.....I.....
	Serca2b	241	...ER.....I...I.....I.....
	Serca3a	241	.V.PER...R...R...HA..V....V....A..A....L..V.....
	Serca3b	241	.V.PER...R...R...HA..V....V....A..A....L..V.....
	Serca3c	241	.V.PER...R...R...HA..V....V....A..A....L..V.....
40	Serca1a	301	ALAVAAIPEGLPAVITTCLALGTRMAKKNAIVRSLPSVETLGCTSVICSDKTGTLTTNQ
	Serca2a	301	.....
	Serca2b	301	.....
	Serca3a	301	.....R.....
	Serca3b	301	.....R.....
45	Serca3c	301	.....R.....
	Serca1a	361	MSVCKMFIIDKVDGDVCSLNEFSITGSTYAPEGEVLKNDKPVRAGQYDGLVELATICALC
	Serca2a	361	...R...L...E..T.....I...Q.D...KCH.....
	Serca2b	361	...R...L...E..T.....I...Q.D...KCH.....
50	Serca3a	361	...R...VVAEAEAGT.R.H..T.S.T..T....RQGEQ...C..F.....
	Serca3b	361	...R...VVAEAEAGT.R.H..T.S.T..T....RQGEQ...C..F.....
	Serca3c	361	...R...VVAEAEAGT.R.H..T.S.T..T....RQGEQ...C..F.....
	Serca1a	421	NDSSLDNFNETKGVYEKVGGEATETALTTLVEKMNVFNTEVRSLSKVERANACNSVIRQLMK
55	Serca2a	421	...A..Y..A.....C.....D..LKG...I.....K....
	Serca2b	421	...A..Y..A.....C.....D..LKG...I.....K....
	Serca3a	421	...A..Y..A.....C.....D..DLKG..R....G.....K...R
	Serca3b	421	...A..Y..A.....C.....D..DLKG..R....G.....K...R
	Serca3c	421	...A..Y..A.....C.....D..DLKG..R....G.....K...R

Serca1a 481 KEFTLEFSRDRKSM SVYCSPAKSSRAAVGNKMFVKGAPEGVIDRCNYVRVGTTTRVPLTGP  
Serca2a 481 .....T.N.P..TSMS-.....THI...S.K..M.PG  
Serca2b 481 .....T.N.P..TSMS-.....THI...S.K..M.PG  
Serca3a 481 .....T.TRADPKVQ.S.....S..E..SS....SRTA..STT  
5 Serca3b 481 .....T.TRADPKVQ.S.....S..E..SS....SRTA..STT  
Serca3c 481 .....T.TRADPKVQ.S.....S..E..SS....SRTA..STT

Serca1a 541 VKEKIMSVIKEWGTGRDTLRLCLALATRDTPPKREEMVLDDSAKFMEYEMDLTFVGVVGM  
Serca2a 540 ..Q.....R...S.S.....H.N.L....H.E...N.IK..TN....C....  
10 Serca2b 540 ..Q.....R...S.S.....H.N.L....H.E...N.IK..TN....C....  
Serca3a 541 SR.H.LAK.RD..S.S.....RK.D.H...CSR.VQ..T.....C....  
Serca3b 541 SR.H.LAK.RD..S.S.....RK.D.H...CSR.VQ..T.....C....  
Serca3c 541 SR.H.LAK.RD..S.S.....RK.D.H...CSR.VQ..T.....C....

Serca1a 601 DPPRKEVTGSIQLCRDAGIRVIMITGDNKGTAIAICRRIGIFSENEEVTDRAYTGREFDD  
Serca2a 600 ....I..AS.VK...Q.....V.....GQD.D..SK.F.....E  
Serca2b 600 ....I..AS.VK...Q.....V.....GQD.D..SK.F.....E  
Serca3a 601 ....P..AAC.TR.SR....V.....V....L...GDT.D.LGK.....  
Serca3b 601 ....P..AAC.TR.SR....V.....V....L...GDT.D.LGK.....  
20 Serca3c 601 ....P..AAC.TR.SR....V.....V....L...GDT.D.LGK.....

Serca1a 661 LPLAEQREACRRACCFARVEPSHKSKIVEYLQSYDEITAMTGDGVNDAPALKKAEIGIAM  
Serca2a 660 .SPSA..D..LN.R.....F...F.....S.....  
Serca2b 660 .SPSA..D..LN.R.....F...F.....S.....  
25 Serca3a 661 .SPEQ..Q...T.R.....A...R...N...FN.....  
Serca3b 661 .SPEQ..Q...T.R.....A...R...N...FN.....  
Serca3c 661 .SPEQ..Q...T.R.....A...R...N...FN.....

Serca1a 721 GSGTAVAKTASEMVLADDNFSTIVAAVEEGRAIYNNMKQFIRYLISSNVGEVVCIFLTAA  
30 Serca2a 720 .....  
Serca2b 720 .....  
Serca3a 721 .....S.A...S...AS.....I  
Serca3b 721 .....S.A...S...AS.....I  
Serca3c 721 .....S.A...S...AS.....I  
35

Serca1a 781 LGLPEALIPVQLLWVNLVTDGLPATALGFNPPDLDIMDRPPRSPKEPLISGWLFFRYMAI  
Serca2a 780 ..F.....NK...N.....L..  
Serca2b 780 ..F.....NK...N.....L..  
Serca3a 781 .....EK...N.R.A.....L..  
40 Serca3b 781 .....EK...N.R.A.....L..  
Serca3c 781 .....EK...N.R.A.....L..  
35

Serca1a 841 GGYVGAATVGAAAWFLYAEDGPHVSYHQLTHFMQCTEHNPEFDGLDCEVFEAPEPMTMA  
Serca2a 840 .C.....IA.DG..R..FY..S..L..K.D..D...V..AI..S.Y....  
45 Serca2b 840 .C.....IA.DG..R..FY..S..L..K.D..D...V..AI..S.Y....  
Serca3a 841 .V...L...A..T....DAE..Q.TFY..RN.LK.S.D..L.A.I..K...SRF.T...  
Serca3b 841 .V...L...A..T....DAE..Q.TFY..RN.LK.S.D..L.A.I..K...SRF.T...  
Serca3c 841 .V...L...A..T....DAE..Q.TFY..RN.LK.S.D..L.A.I..K...SRF.T...

Serca1a 901 LSVLVTIEMCNALNSLSENQSLLRMPWPVNIWLLGSICLSMSLHFLILYVDPLPMIFKLR  
Serca2a 900 .....E...V.....E...L...QIT  
Serca2b 900 .....E...V.....E...L...QIT  
Serca3a 901 .....V.....L.P....AVVM..A.....L.P...L...QVT  
Serca3b 901 .....V.....L.P....AVVM..A.....L.P...L...QVT  
55 Serca3c 901 .....V.....L.P....AVVM..A.....L.P...L...QVT

Serca1a 961 ALDFTQWLMVLKISLPVIGLDELLKFIARNYLEG  
Serca2a 960 P.NL.....LM..T...V.....QPAILE  
Serca2b 960 P.NL.....LM..T...V.....QPGKECVQPATKSSCSLSACTDGISWP  
Serca3a 961 P.SGR..GV..QM.....L...A..YLS..HMDEKKDLK  
5 Serca3b 961 P.SGR..GV..QM.....L...A..YLS..HMD.VLGTFMQARSRLPTTSRTPYHTGKK  
Serca3c 961 P.SGR..GV..QM.....L...A..YLS..HMD.VLGTFMQARSRLPTTSRTPYHTGLA

Serca2b 1020 FVLLIMPLVVVVYSTDTNFSDFWWS  
10 Serca3b 1021 GPEVNPGSRGESPVWPSD  
Serca3c 1021 SWKKRT

**Figure 2****Sequence similarity of Serca2 proteins in mammalian species**

	Mouse_2a	1	MENAHTKTVEEVLGHFGVNESTGLSLEQVKKLKERWGSNELPAEEGKTLLLELVIEQFEDL
5	Mouse_2b	1	.....
	Rat_2b	1	.....
	Rat_2a	1	.....
	Dog_2a	1	.....
	Cat_2a	1	.....Y.....
10	Pig_2a	1	.....
	Pig_2b	1	.....
	Human_2b	1	.....
	Human_2c	1	.....
	Human_2a	1	.....
15	Rabbit_2a	1	.....
	Rabbit_2b	1	.....
	Mouse_2a	61	LVRILLLAACISFVLAWFEEGEETITAFVEPFVILLILVANAIVGVWQERNAENAIEALK
	Mouse_2b	61	.....
20	Rat_2b	61	.....
	Rat_2a	61	.....
	Dog_2a	61	.....
	Cat_2a	61	.....
	Pig_2a	61	.....
25	Pig_2b	61	.....
	Human_2b	61	.....
	Human_2c	61	.....
	Human_2a	61	.....
30	Rabbit_2a	61	.....
	Rabbit_2b	61	.....
	Mouse_2a	121	EYEPENGKVYRQDRKSVQRIKAKDIVPGDIVEIAVGDKVPADIRLTSIKSTTLRVDQSIL
	Mouse_2b	121	.....
	Rat_2b	121	.....
35	Rat_2a	121	.....
	Dog_2a	121	.....
	Cat_2a	121	.....
	Pig_2a	121	.....
	Pig_2b	121	.....
40	Human_2b	121	.....
	Human_2c	121	.....
	Human_2a	121	.....
	Rabbit_2a	121	.....
	Rabbit_2b	121	.....
45	Mouse_2a	181	TGESVSVIKHTDPVPDPRAVNQDKKNMLFSGTNIAAGKAMGVVVATGVNTEIGKIRDEM
	Mouse_2b	181	.....
	Rat_2b	181	.....
	Rat_2a	181	.....
50	Dog_2a	181	.....
	Cat_2a	181	.....
	Pig_2a	181	.....
	Pig_2b	181	.....
	Human_2b	181	.....
55	Human_2c	181	.....
	Human_2a	181	.....
	Rabbit_2a	181	.....
	Rabbit_2b	181	.....

	Mouse_2a	241	ATEQERTPLQQKLDEFGEQLSKVISLICIAVWIINIGHFNDPVGGSWIRGAIYYFKIAV
	Mouse_2b	241	.....
	Rat_2b	241	.....
5	Rat_2a	241	.....
	Dog_2a	241	.....
	Cat_2a	241	.....
	Pig_2a	241	.....
	Pig_2b	241	.....
10	Human_2b	241	.....
	Human_2c	241	.....
	Human_2a	241	.....
	Rabbit_2a	241	.....
	Rabbit_2b	241	.....
15	Mouse_2a	301	ALAVAAIPEGLPAVITTCLALGTRRMAKKNAIVRSLPSVETLGCTSVICSDKTGTLTTNQ
	Mouse_2b	301	.....
	Rat_2b	301	.....
	Rat_2a	301	.....
20	Dog_2a	301	.....
	Cat_2a	301	.....
	Pig_2a	301	.....
	Pig_2b	301	.....
	Human_2b	301	.....
25	Human_2c	301	.....
	Human_2a	301	.....
	Rabbit_2a	301	.....
	Rabbit_2b	301	.....
30	Mouse_2a	361	MSVCRMFI LDKVEGDTCSLNEFSITGSTYAPIGEVQKDDKPVKCHQYDGLVELATICALC
	Mouse_2b	361	.....
	Rat_2b	361	.....T.....
	Rat_2a	361	.....T.....
	Dog_2a	361	.....R...S...T.....H.....
35	Cat_2a	361	.....T.....H.....
	Pig_2a	361	.....T.....H.....
	Pig_2b	361	.....T.....H.....
	Human_2b	361	.....R.....T.....H.....N.....
	Human_2c	361	.....R.....T.....H.....N.....
40	Human_2a	361	.....R.....T.....H.....N.....
	Rabbit_2a	361	.....D.....T.....H.....
	Rabbit_2b	361	.....D.....T.....H.....
45	Mouse_2a	421	NDSALDYNEAKGVYEKVGEATETALTCLVEKMNVFDTELKGLSKIERANACNSVIKQLMK
	Mouse_2b	421	.....
	Rat_2b	421	.....
	Rat_2a	421	.....
	Dog_2a	421	.....
	Cat_2a	421	.....K.F.....
50	Pig_2a	421	.....
	Pig_2b	421	.....
	Human_2b	421	.....
	Human_2c	421	.....
	Human_2a	421	.....
55	Rabbit_2a	421	.....
	Rabbit_2b	421	.....

	Mouse_2a	481	KEFTLEFSRDRKSMSVYCTPNKPSRTSMSKMFVKGAPEGVIDRCTHIRVGSTKVPMTPGV	
	Mouse_2b	481	.....	
	Rat_2b	481	.....	
	Rat_2a	481	.....	
5	Dog_2a	481	.....	
	Cat_2a	481	.....	
	Pig_2a	481	.....	
	Pig_2b	481	.....	
	Human_2b	481	.....	S.
10	Human_2c	481	.....	S.
	Human_2a	481	.....	S.
	Rabbit_2a	481	.....	A.
	Rabbit_2b	481	.....	A.
15	Mouse_2a	541	KQKIMSVIREWGSGSDTLRCLALATHDNPLKREEMHLEDSANFIKYETNLTFVGCVGMLD	
	Mouse_2b	541	.....	
	Rat_2b	541	.....	R.
	Rat_2a	541	.....	R.
	Dog_2a	541	...V.....	R...N
20	Cat_2a	541	...V.....	R...N
	Pig_2a	541	.....	MR...N
	Pig_2b	541	.....	MR...N
	Human_2b	541	.....	R.
	Human_2c	541	.....	R.
25	Human_2a	541	.....	R.
	Rabbit_2a	541	.....	R...K
	Rabbit_2b	541	.....	R...K
30	Mouse_2a	601	PPRIEVASSVKLCRQAGIRVIMITGDNKGTAVAICRRIGIFGQDEDVTSKAFTGREFDEL	
	Mouse_2b	601	.....	
	Rat_2b	601	.....	
	Rat_2a	601	.....	
	Dog_2a	601	.....	
	Cat_2a	601	.....	
35	Pig_2a	601	.....	
	Pig_2b	601	.....	
	Human_2b	601	.....	
	Human_2c	601	.....	
	Human_2a	601	.....	
40	Rabbit_2a	601	.....	E...A
	Rabbit_2b	601	.....	E...A
45	Mouse_2a	661	SPSAQRDACLNARCFARVEPSHKSKIVEFLQSFDEITAMTGDGVNDAPALKKSEIGIAMG	
	Mouse_2b	661	.....	
	Rat_2b	661	.....	
	Rat_2a	661	.....	
	Dog_2a	661	.....	
	Cat_2a	661	.....	
	Pig_2a	661	N...E.....	
50	Pig_2b	661	N...E.....	
	Human_2b	661	N.....	A.
	Human_2c	661	N.....	A.
	Human_2a	661	N.....	A.
	Rabbit_2a	661	N.....	A.
55	Rabbit_2b	661	N.....	A.

	Mouse_2a	721	SGTAVAKTASEMVLADDNFSTIVAAVEEGRAIYNNMKQFIRYLISSNVGEVVCIFLTAAL
	Mouse_2b	721	.....
	Rat_2b	721	.....
	Rat_2a	721	.....
5	Dog_2a	721	.....
	Cat_2a	721	.....
	Pig_2a	721	.....
	Pig_2b	721	.....
	Human_2b	721	.....
10	Human_2c	721	.....
	Human_2a	721	.....
	Rabbit_2a	721	.....
	Rabbit_2b	721	.....
15	Mouse_2a	781	GFPEALIPVQLLWVNLVTDGLPATALGFNPDDLDMNKPPRNPKEPLISGWLFFRYLAIG
	Mouse_2b	781	.....
	Rat_2b	781	.....
	Rat_2a	781	.....
	Dog_2a	781	.....
20	Cat_2a	781	.....
	Pig_2a	781	.....
	Pig_2b	781	.....
	Human_2b	781	.....
	Human_2c	781	.....
25	Human_2a	781	.....
	Rabbit_2a	781	.....
	Rabbit_2b	781	.....
30	Mouse_2a	841	CYVGAATVGAAAWWFIAADGGPRVSFYQLSHFLQCKEDNPDFDGVDCAI FESPYPMTMAL
	Mouse_2b	841	.....
	Rat_2b	841	.....E.....
	Rat_2a	841	.....E.....
	Dog_2a	841	.....D.....E.....
	Cat_2a	841	.....D.....E.....
35	Pig_2a	841	.....T.....E.....V.....
	Pig_2b	841	.....T.....E.....V.....
	Human_2b	841	.....E.....
	Human_2c	841	.....E.....
	Human_2a	841	.....E.....
40	Rabbit_2a	841	.....E.....
	Rabbit_2b	841	.....E.....
45	Mouse_2a	901	SVLVTTIEMCNALNSLSENQSLLRMPWENIWLVGSI CLSMLS LHFLILYVEPLPLIFQITP
	Mouse_2b	901	.....
	Rat_2b	901	.....
	Rat_2a	901	.....
	Dog_2a	901	.....
	Cat_2a	901	.....
	Pig_2a	901	.....
50	Pig_2b	901	.....
	Human_2b	901	.....
	Human_2c	901	.....
	Human_2a	901	.....
	Rabbit_2a	901	.....
55	Rabbit_2b	901	.....

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Mouse_2a  961  LNLQWLMVLKISLPVILMDETLKFVARNYLEQPAILE-----
Mouse_2b  961  .....GKECVQPATKSSCSLSACTDGISWPF
Rat_2b    961  .....-GKECA.....P.....
Rat_2a    961  .....-AILE
5  Dog_2a  961  .....-AILE
   Cat_2a  961  .....-AILE
   Pig_2a  961  .....-AILE
   Pig_2b  961  .....-GKEC.....-F.....
   Human_2b 961  ..V.....-GKEC.....-F.....
10  Human_2c 961  ..V.....-VLSSL
   Human_2a 961  ..V.....-AILE
   Rabbit_2a 961  ..V.....-AILE
   Rabbit_2b 961  ..V.....-GKEC....PQ....W...E.V....

15  Mouse_2b 1021  VLLIMPLVVVVYSTDTNFSDMFWS
   Rat_2b    1020  .....
   Pig_2b    1019  .....
   Human_2b  1019  .....
   Rabbit_2b 1019  ....V...M.....LL..

20
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**Fig. 3 Targeting construct for Serca2 flox gene modification. Sequence information.**

----- Serca2 gene -----

**LoxP site 1: Intron 1**  
 underlined sequence = loxP site and cloning sequence

**Exon I (partial)**

CACGGGGCTGAGCTTGGAGCAGGTCAAGAAGCTCAAGGAGAGATGGGGCTCCAACGgtaggtgcggggcg  
 ccggggctgcagcggcgcgggcgggcccgagcgccaaggaagatgggtgaccgggtccacctcgtggg  
 gcttggctcggcgcgggcgcccgaaggctgcgagaggccggcggtccaagcgcggggtctggggccatcgccg  
 10 accttaggggtctcgaatcaagcttatcgataccgtcgatcggaacctcgagggggggcccggtacccggg  
gatcaattcgagctcgcccggggatcgatccggaaccttaataATAACTTCGTATAATGTATGCTATACG  
AAGTTATTtaggtccctcgacctgcagcccaagctccGGGGATctcgagccgggtgaccttcccgccggcg  
 ctgagcgagtcggattgggggggggggagagggagtgaggaggaggaggttccctgcggctgggctg  
 agtcccccgcggtattatgaggcgctgatgttggtagaaacctcggaacctttcttgtgctcccaaa  
 15 gttgcacatctggcagaagtgatgaccagctgaaatgactgcatggctctggaggccggagagggctta  
 cgggcagttccgaggccactgattaccagggctgaataattttctcggggtatcaaagtggagacagatt  
 gttgtacgttcatacacctatatccgccattcagacaacgatgggtggtgaatttagcagtttttaataaa  
 agcgctaatacaatatcttcattttttctttc

----- Serca2 gene -----

**LoxP site 2: Intron 3 5' of genomic XcmI site**  
 underlined sequence = loxP site, cloning sites and partial HSV-TK

ccaaatTTTTattcttagaacaattgtattcttatactgtgtaggaagtgaataatcatacagtacttgtc  
 ttaggtttcacaataactgataactgtatgggtttcaattatgtattcacacgtttaagtctgacccaggggG  
 GATCCggaaccttaataATAACTTCGTATAATGTATGCTATACGAAGTTATTtaggtccctcgacctgcag  
 25 cccaagctgatccctctagtcgagcccgagctgggttctttccgcctcagaagccatagagccaccgcac  
cccagcatgcctgctattgtcttcccaatccctcccccttgctgtcctgccccacccccacccccagaata  
gaatgacacctactcagacaatgcgatgcaatttctctcattttatttaggaaaggacagtgaggagtgccac  
cttccagggtcaaggaaggcacgggggaggggcaacaacagatggctggcaactagaaggcacagtcga  
ggctgatcagcgagctctagctagagaattgatccccctcagaagaactcgtcaagaaggcgatagaaggc  
 30 gatgcgctgcgaatcgggagcgccgat\*ccgtaaagcacgaggaagcgg\*cgcccatcgcgcgccaagc  
tctttcagcaatatcacgggtagccaacgctatgt\*ctgataagcgggtccgcccacacccaa\*ccggcca  
caagtc\*atgaaatcca\*aaaaagcgggcccatttttccacc\*atgatttt\*cggaagcaaggccctt\*c  
cattgggtcaccgac\*aga\*catt\*tcggt\*c\*ggcattgcgc\*ccct

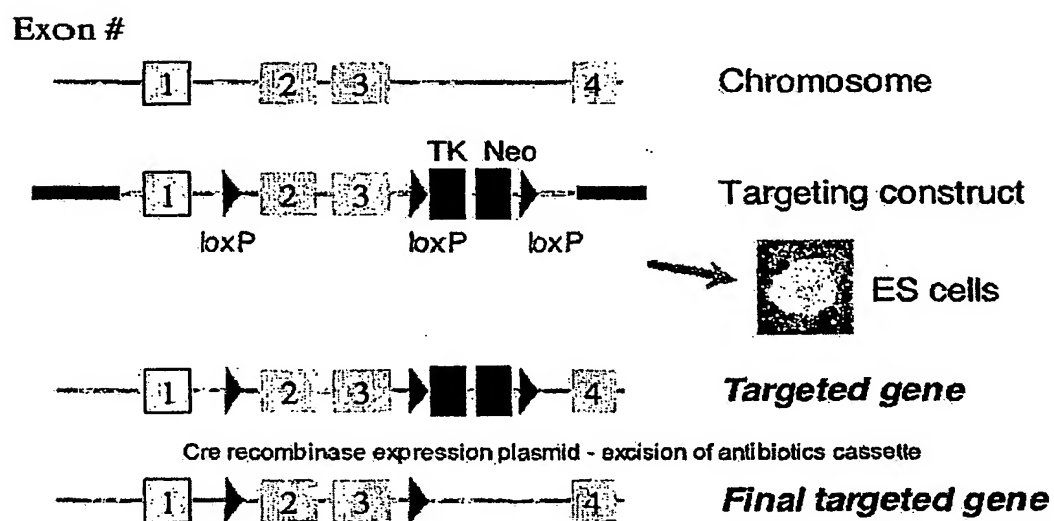
----- HSV-TK Neo antibiotics cassette -----

**LoxP site 3: Intron 3 5' of genomic XcmI site**  
 underlined sequence = loxP site, cloning sites and partial Neo gene

gttttcat\*accaccgcggggtcccgggc\*gatat\*ttcaccttgct\*ag\*cggtgttggtgtggtgtaaatg  
 ttgcgcatgttttcgaaagcccc\*agcaccgcccagtaagtcacggctcggttacgtagacgatatcgtc  
 gcgcgaacccagggccaccagcaagttgcgtgggtgggttttccccatcc\*gtggggac\*gtctatataa  
 40 acc\*gcagtagcgtgggcattttctgctcggggcggaactccgctggcttcttgcctgcggcgagggcgcaa  
cgccgtacgtcggttgctatggccgcgagaacgcgcagcctgggtcgaacgcagacgcgtgttgatggccgg  
ggtacgaagccatacgcgcttctacaaggcgctggccgaagaggtgcgggagtttcacgcccaccaagatct  
gcggcacgctgttgacgctgttaagcgggtcgctgcagggtcgctcggtgttcgaggccacacgcgtcacc  
ttaatatgcgaagtggacctcggaacgcgcgccccgactgcattctgcgtgttcgaattcgccaatgacaa  
 45 gacgctggggcggggtttgctcgacattgggtggaaacattccaggcctgggtggagagggcttttgccttcc  
tcttgcaaaaccacactgctcgacattgggtggaaacattccaggcctgggtggagagggcttttgccttcc  
tcttgcaaaaccacactgctcgatccggaaccttaataATAACTTCGTATAATGTATGCTATACGAA  
GTTATTtaggtccctcgacctgcagcccaagctgatcctctagaqtcgacctcgatctggtggtcatggccctatgaaa  
acattagcttagagg

**Fig. 4A** Schematic representation of genetic manipulation.

### Serca2 (*atp2a2*) gene modification



**Fig 4 B: Verification of Serca locus targeting events offspring from chimeric mice.**

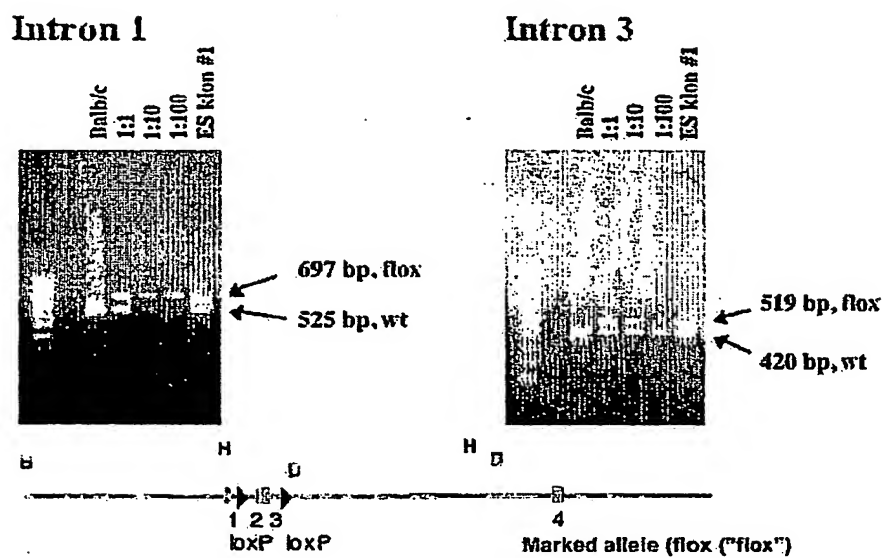


Fig. 5 Specificity of gene deletion in a test model.

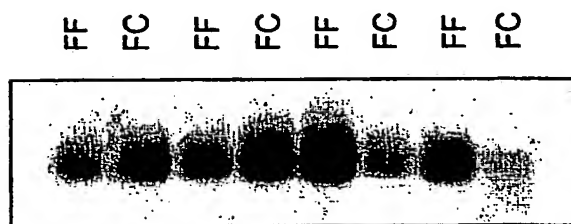


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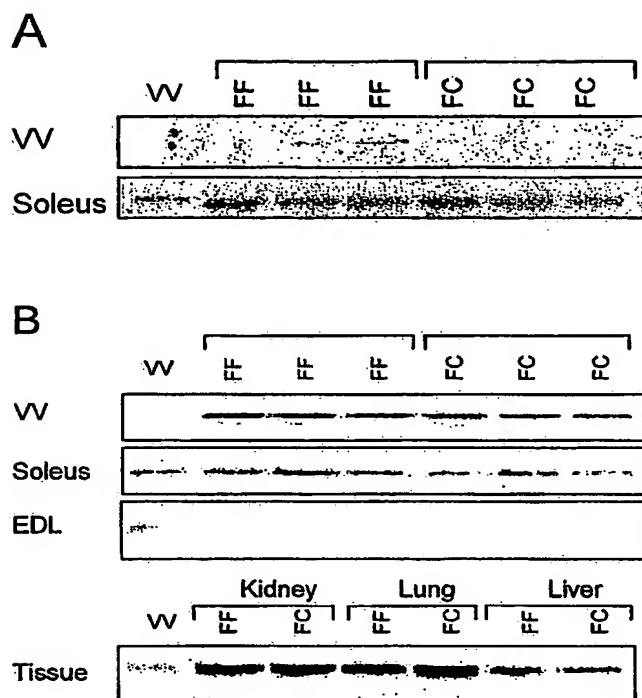
- 10 Control A = wt ES cells (14.1a)  
 Control B = Upper panel: Serca2<sup>wt/flox</sup> ES cells  
 Middle panel: Left ventricle from MLC-2V-Cre mice  
 Lower panel: Serca2<sup>wt/del</sup> ES cells
- 15 LV = heart left ventricle  
 RV = heart right ventricle  
 EDL = extensor digitorum longus muscle (fast-twitch skeletal muscle)  
 Soleus = soleus muscle (slow-twitch skeletal muscle)  
 Other tissues as indicated.

**Fig. 6 Cardiac ANP mRNA expression.**

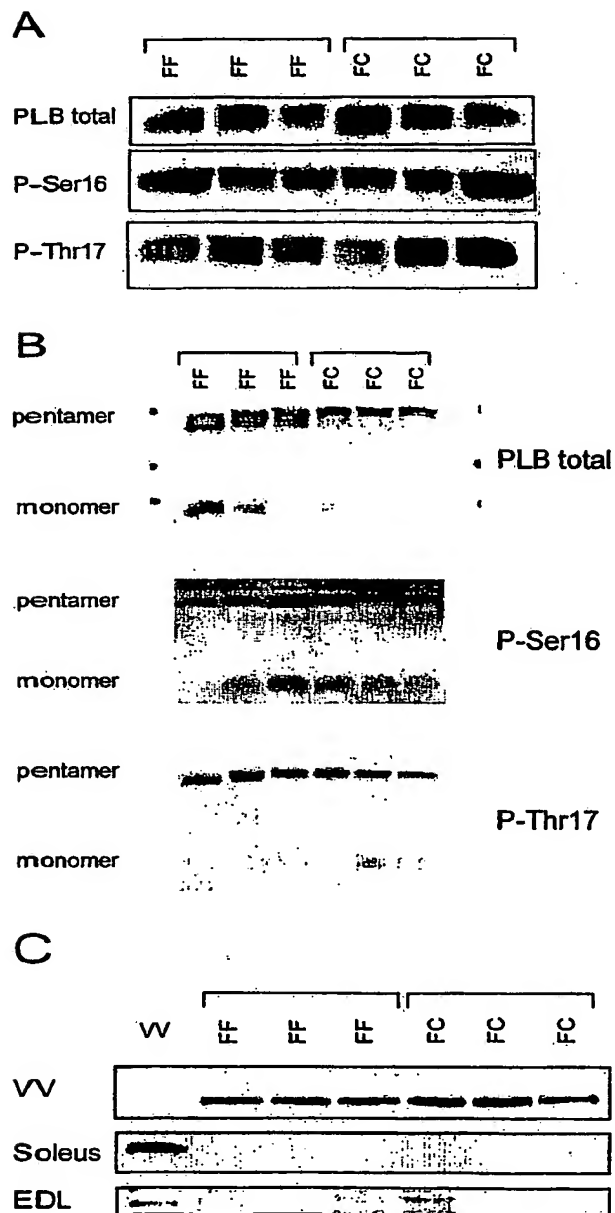
5



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**Fig. 7 Serca2 protein expression.**

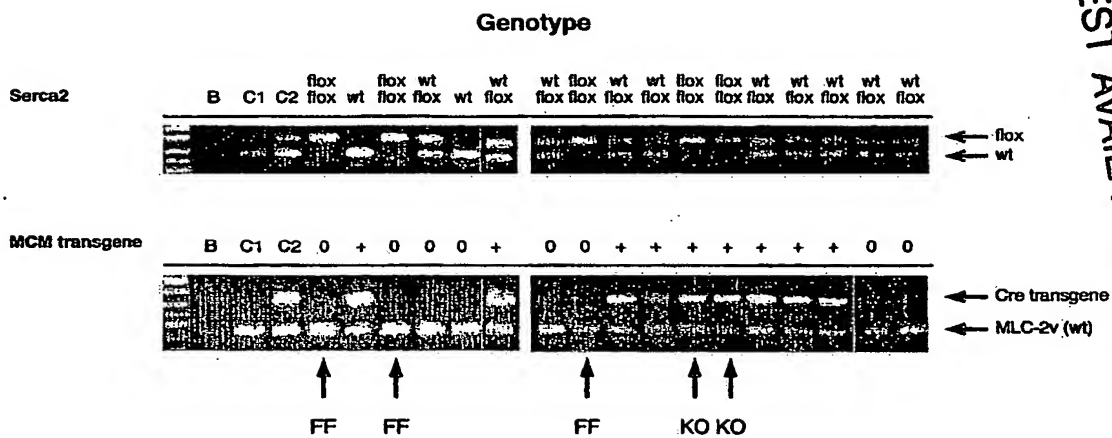
**Fig. 8 Compensatory mechanisms in *Serca<sup>fllox</sup>* MLC-2 $\nu$ -Cre mice.**



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**Figure 9 Genotypes PCR**

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10 Generation of animals with Serca2<sup>flox</sup> and MCM transgene alleles.

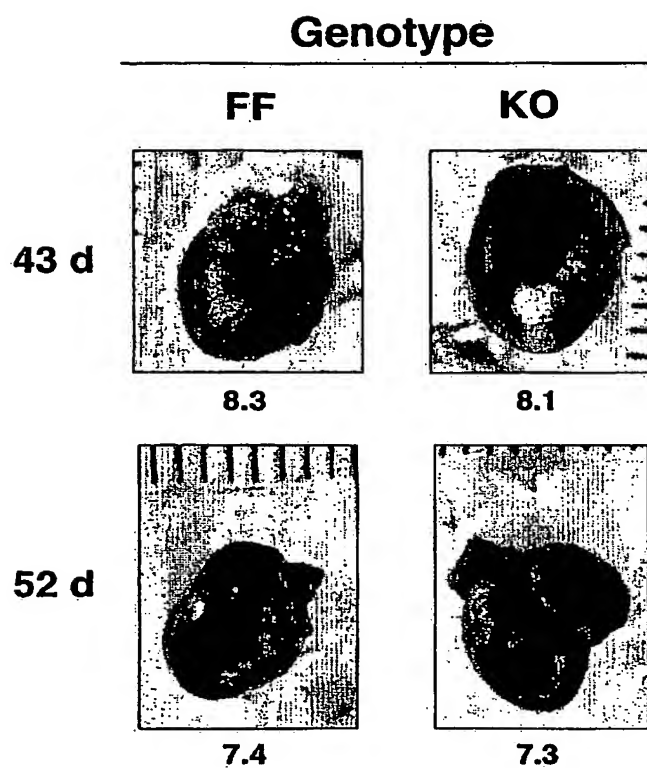
Genotypes FF. Serca2<sup>flox/flox</sup>; KO, Serca2<sup>flox/flox</sup> MCM



Figure 10

## Heart morphology

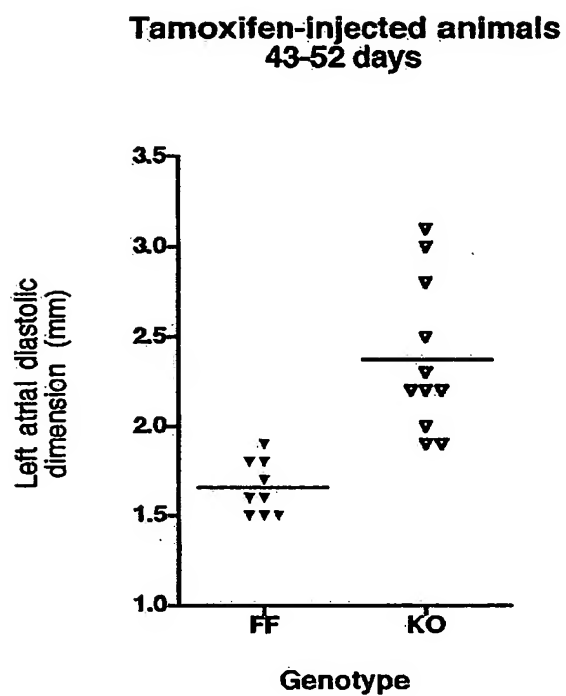
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**Figure 11 Pilot series left atrial diastolic diameter.**

5

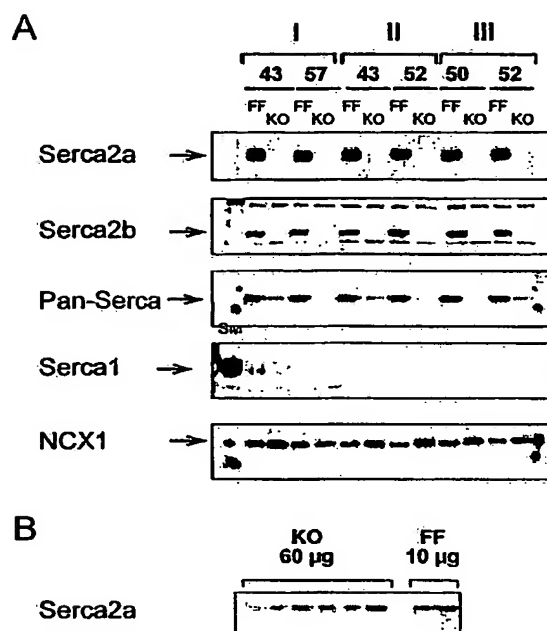


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**Figure 12 Serca protein content in tamoxifen-induced FF and KO mice**

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